REMARKS

Upon entry of this response, claims 13-19 and 26-28 will remain pending, with claims 13, 15 and 17-19 being independent claims.

Reconsideration and allowance of the application are respectfully requested.

Response To Rejection

The following rejection is set forth in the Office Action:

Claims 13-19 and 26-28 are rejected under 35 U.S.C. 102(b) as being anticipated by Engman et al. (hereinafter "Engman"), "Diaryl Chalcogenides as Selective Inhibitors of Thioredoxin Reductase and Potential Antitumor Agents", Anticancer Research, Helenic Anticancer Institute, Anthens, GR, Vol. 17, No. 6D, 1997, pp. 4599-4605.

Applicants submit that the rejection is without appropriate basis in that (as previously argued by Applicants throughout the lengthy prosecution of this application) Engman discloses that ebselen was found to be an inhibitor of human thioredoxin reductase. Engman does not disclose the use of ebselen as a substrate for thioredoxin reductase. See, for example, the abstract of Engman. Engman merely concludes that ebselen is an inhibitor of thioredoxin reductase. As stated in Engman's abstract, ebselen is not a competitive inhibitor for thioredoxin, but for thioredoxin reductase.

With regard to the above, Applicants respectfully point out that the Final Office Action includes a number of misstatements in support of the rejection. Therefore, reconsideration of Applicants' claimed subject matter with correction of the misstatements is requested along with clarification of the rejection, if the rejection is maintained. In this regard, because the rejection is unclear, especially in view of misstatements in the Final Office Action, Applicants submit that the

rejection should be withdrawn, and the application allowed. Moreover, if the rejection is clarified, the finality of the Office Action should be withdrawn.

In particular, the Final Office Action makes an assertion, on page 4, in Response to Arguments, that "Please note that a competitive inhibitor is also a substrate." However, substrates and competitive inhibitors are not the same. In fact, a competitive inhibitor competes with a substrate, and the two are mutually exclusive. See, for example, "Competitive Inhibitors" (which includes animated graphics that do not clearly show in a print out), downloaded March 18, 2009 from

http://www-biol.paisley.ac.uk/Kinctics/Chapter_3/chapter3_2.html and "Competitive Inhibition" downloaded March 18, 2009 from

http://en.wikipedia.org/wiki/Compctitive_inhibitor.

Moreover, the Final Office Action, at the same location, contends that selenite is indicated to be a substrate in Engman. However, there is no correlation provided in the Final Office Action as to why ebselen can be considered based upon the disclosure of Engman to be a substrate, especially when Engman discloses that ebselen is an inhibitor.

On the top of page 5 of the Final Office Action, it is asserted that, "More the substrate more production of reduced thioredoxin, which in turn enhances the peroxidase activity." There is no support provided for this assertion. Moreover, it is not clear what is intended by this assertion. Therefore, if this assertion is maintained, the Examiner is requested to provide support. Also, clarification is requested because it appears that the affect would be the opposite, with the result being the oxidation of reduced thioredoxin.

Moreover, the further remarks regarding claims 15 and 16 are not clear. According to Applicants' findings, ebselen is reduced by the enzyme or thioredoxin, and the thioredoxin stimulates reduction of ebselen. There does not appear to be any teaching or suggestion in Engman of Applicants' finding.

Regarding claims 17 and 18, the Response to Arguments on page 5 is also unclear. The response contends that, "Thioredoxins are electron donors." However, it is the enzyme thioreductase that is the electron donor. Moreover, in contrast to the assertion in the Office Action, Km for thioredoxin and the selenium compound are the same.

Still further, it is contended in the Final Office Action that "...the unreacted selenium compound expected to oxidize the reduced thioredoxin." However, it is not expected because the selenium compound is disclosed in Engman as being an inhibitor.

Again, Applicants point out that Engman was looking at ebselen as an inhibitor, and used conditions wherein ebselen is used as an inhibitor and not as a substrate. Accordingly, Engman does not disclose each and every feature of Applicants' claims including the conditions recited in Applicants' claims. In particular, Engman discloses in his assay, at page 4600, the paragraph bridging the right and left-hand columns, that, "Thioredoxin reductase activity was measured spectrophotometrically at room temperature by the oxidation of NADPH at 339 nm in the presence of 15μM human recombinant thioredoxin and 1 mg/ml bovine insulin." (Emphasis added.)

Thus, in each of the assays of Engman, insulin is present. Insulin in the assay affects the results, and is included in the assay apparently because Engman was experimenting with ebselen as an inhibitor. The conditions used by Engman are not conditions as recited in Applicants' claims that achieve the results recited in Applicants' claims.

Therefore, Engman does not teach each and every feature recited in Applicants' claims. Engman does not constitute anticipatory prior art as asserted in the rejection, because:

(1) Engman does not disclose, as recited in Applicants' independent claim 13, a method for reduction of a substrate with thioredoxin reductase, comprising combining the thioredoxin reductase, the substrate and NADPH *in vitro* under conditions to reduce the substrate, the substrate being as recited in Applicants' claim 13.

(2) Engman does not disclose, as recited in Applicants' independent claim 15, a method of enhancing peroxidase activity of thioredoxin reductase, comprising combining NAPDH, thioredoxin reductase, thioredoxin and a substrate *in vitro* under conditions to enhance peroxidase activity of thioredoxin reductase, the substrate being as recited in Applicants' claim 15.

(3) Engman does not disclose, as recited in Applicants' independent claim 17, a method of oxidizing reduced thioredoxin by a substrate, the method comprising combining reduced thioredoxin and a substrate *in vitro* under conditions to oxidize the reduced thioredoxin with the substrate, the substrate being as recited in Applicants' claim 17.

(4) Engman does not disclose, as recited in Applicants' independent claim 18, a method for reducing a peroxide comprising combining thioredoxin, thioredoxin reductase, NAPDH and a substrate *in vitro* under conditions to reduce the peroxide, the substrate being as recited in Applicants' claim 18.

(5) Engman does not disclose, as recited in Applicants' independent claim 19, a method of preventing peroxidation of a substance comprising combining thioredoxin, thioredoxin reductase and NADPH with a substrate *in vitro* under conditions to prevent peroxidation of the substance, the substrate being as recited in Applicants' claim 19.

The Examiner is reminded that in contrast to the prior art of record, the present invention recognizes and demonstrates that ebselen is a substrate being reduced by NADPH and thioredoxin reductase with a low Km-value meaning that it is a very good substrate undergoing unlimited cycles of oxidation/reduction in the presence of hydrogen peroxide without affecting the activity of the enzyme. The reduced ebselen is called ebselen selenol and has the Se-N bond broken by reduction. The selenol is oxidized back to ebselen by hydrogen peroxide or another peroxide and a new cycle starts. The reaction is ultimately driven by NADPH. Reduced thioredoxin strongly enhances the thioredoxin reductase reaction which is also proven by determination of the rate of reduction of ebselen by reduced thioredoxin using kinetics with tryptophan fluorescence. The result, never seen before, is that ebselen is a very efficient oxidant of reduced thioredoxin.

Accordingly, for at least the reasons set forth above, each of the pending claims is patentable over Engman, and the rejection should be withdrawn.

CONCLUSION

In view of the foregoing, the Examiner is respectfully requested to reconsider and withdraw the rejection of record, and allow each of the pending claims.

Applicants therefore respectfully request that an early indication of allowance of the application be indicated by the mailing of the Notices of Allowance and Allowability.

Should the Examiner have any questions regarding this application, the Examiner is invited to contact the undersigned at the below-listed telephone number.

Respectfully submitted,
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